



Influence of Dietary Inclusion of Sunflower (*Helianthus Annus*) Oil on Growth Performance and Oxidative Status of Broiler Chicks

1. Agubosi O.C.P.
2. Alexander James
3. Alagbe J.O

Received 25th Dec 2021,
Accepted 30th Jan 2022,
Online 24th Feb 2022

^{1,2,3} Department of Animal Science,
University of Abuja, Nigeria
Agubosi.oluchi@uniabuja.edu.ng

Abstract: A study was conducted to evaluate the dietary inclusion levels of Sunflower oil (SFO) on the growth performance and oxidative status of broiler chicks. A total of one hundred and fifty (150) day old Abor acre chicks were randomly allotted into five dietary treatments in a completely randomized design (CRD). Each treatment was replicated thrice with ten birds per replicate. A basal diet was formulated to meet the nutrient requirement of broiler chicks. Birds on treatments 2, 3, 4 and 5 were fed the basal diet mixed with 0.1, 0.2, 0.3 and 0.4 mls of sunflower oil, respectively while birds on treatment 1 was fed the basal diet with Oxytetracycline at 1.2g/kg. Feed and clean water were provided ad libitum throughout the experimental period which lasted for 28 days. Data were collected on average daily feed intake (ADFI), average daily weight gain (ADWG), feed conversion ratio (FCR) and oxidative status. Results on phytochemical composition of sunflower oil revealed the presence of alkaloid (3.86 %), condensed tannins (0.96 %), hydrolysable tannins (9.86 %), phenols (7.06 %), terpenoids (5.10 %), steroids (4.47%), saponins (3.51%) and flavonoids (10.06 %). ADWG, ADFI and mortality values were significantly ($P < 0.05$) different in all the parameters measured.

Keywords: Antibiotics, *Helianthus annus*, Phytochemicals, Performance, Broilers.

Birds fed 0.2, 0.3 and 0.4 mls of sunflower oil had similar final body weight gain, average weight gain and feed conversion ratio which is significantly ($p < 0.05$) higher than those fed 0.1 mls of sunflower oil. Similarly ADFI of birds fed 0.2mls of sunflower oil was significantly ($P < 0.05$) higher than those fed 0.1, 0.3 and 0.4mls. Activities of superoxide dismutase (SDA), glutathione peroxidase (GPx), catalase (CAT) and malonyldialdehyde (MLA) were significantly ($P < 0.05$) influenced by the treatments. It can be concluded that the better performance observed in birds fed sunflower oil could

be attributed to the activities of bioactive chemicals contained in sunflower oil which has been proven to stimulate functions of the intestinal tract to improve digestive secretions and scavenge free radicals.

Introduction

In the recent years, there has been increasing concern over the indiscriminate use of antibiotics in livestock production due to the possible emergence of multi-drug resistance, dangers of environmental pollution and toxic residue in animal products (Diarra and Malouin, 2014; Alagbe, 2019). Consumption of animal products (meat, milk and eggs) with residues of antibiotics can actually have a negative impact on human health (Oluwafemi *et al.*, 2021). Plants produce primary and secondary metabolites which encompasses a whole array of function (Croteau, 2000). Primary metabolites include amino acids, carbohydrates, proteins and lipids, are compounds that are necessary for cellular processes (Chinelo and Ajunwa, 2017; Adewale *et al.*, 2021). Secondary metabolites or phytochemicals include compounds with high therapeutic properties such as: alkaloids, flavonoids, tannins, saponins, phenols, terpenoids and so on (Chiasson *et al.*, 2001; Alagbe and Omokore, 2019).

The use of plant extract has been proven to help prevent the residual effect of synthetic chemicals, via the use of oil from plants. Essential oils (EOs), also called volatile odoriferous oil, are aromatic oily liquids extracted from different parts of plants, for example, leaves, peels, barks, flowers, buds, seeds, and so on (Windisch *et al.*, 2008; Agubosi *et al.*, 2021). EOs from plants can be extracted via several methods such as: steam distillation, soxhlet method, cold press expression and so on (Cassel and Vargas 2006; WHO, 2014).

Sunflower (*Helianthus annuus*) oil has remarkable bactericidal properties as reported by Lezcono *et al.* (1998) in a preliminary study, and acts directly on the pathogenic micro-organism without causing damage. It is an oil-rich crop that is well-known for its nutraceutically important metabolites along with the taste and popularity of the oil (Ngibad, 2019).

Studies have revealed that sunflower seeds are rich nutrients and certain different phytochemicals such as antioxidants, flavonols, phenolic acids, procyanidins, phytosterols, amino acids, dietary fiber, potassium, arginine monounsaturated, and polyunsaturated fatty acids which contribute to the improvement of human health (Alagawany *et al.*, 2015; Islam *et al.*, 2016). The presence of some essential amino acids such as aspartic acid, glutamic acid, serine, histidine, glycine, threonine, arginine, alanine, tyrosine, cysteine, valine, methionine, phenylalanine, isoleucine, leucine, lysine, and proline in sunflower products has also been reported (Karangwa *et al.*, 2015). The nutritional composition of sunflower seeds and oil has dictated their functional properties, also effective in preventing or controlling human diseases such as diabetes, cancers, hypertension, hypercholesterolemia, and coronary heart disease (Katsarou *et al.*, 2015).

Sunflower oil possesses antitumor, anti-inflammatory, antioxidant, skin-protective, anticancer, antimalarial, hypocholesterolemic, antihypertensive, analgesic, and antimicrobial activity and silent effects on muscles, nerves, and blood vessel (Zoumpoulakis *et al.*, 2017) and the stem bark, roots and leaf extract has been used traditionally for the treatment of dysentery, cough, skin rashes, gastrointestinal disorders, diarrhea, sores, etc. (Mohiuddin, 2019).

In view of the abundant potential in Sunflower oil, this experiment was designed to determine the influence of dietary inclusion of sunflower (*Helianthus annuus*) oil on the growth performance and oxidative status of broiler chicks.

Materials and methods

Site of the experiment

This study was carried out at the University of Abuja Teaching and research farm, University of Abuja, which is situated at Gwagwalada area council, F. C. T. Abuja, Nigeria. It falls between latitude 08° 51' and 09° 37'N and longitude of 70° 20' and 07° 51'E.

Collection of plant material and extraction of Sunflower oil

Helianthus annuus seeds were collected within University of Abuja Teaching and Research Farm Abuja and authenticated at the Department of Crop Science, University of Abuja, Gwagwalada, Nigeria where a voucher specimen was deposited with a reference number HA/2ABJM. Seeds were sun dried for 8 days and pulverized into powder using laboratory grinder. 2000 grams of the powdered seeds was put into a porous thimble and placed in a Soxhlet extractor (GH-2A11 model, Punjab, India) using 300 ml of n-Hexane as extracting solvent for 2 hours until the required quantity was obtained, oil was kept in a well labeled container for further analysis.

Animal management and experimental procedure

150-1-day-old Arbor acre broiler chicks of mixed sex were obtained from a reputable commercial hatchery in Ibadan, Oyo state, Nigeria. Two weeks prior to the arrival of the birds, battery cage measuring (550 cm × 200 cm × 100 cm) (L×B×H) were thoroughly washed and disinfected. Birds were given anti-stress (Glucomol® at 10 g to 10 liters of water) and randomly assigned into 5 treatments (designated as T1, T2, T3, T4 and T5) with 3 replicates consisting of 10 birds each in a completely randomized design. Basal diet was formulated to meet the nutrient requirement of broilers according to Aduku (1994). Vaccination program was designed according to the prevailing disease condition of the environment and strict biosecurity measures was put in place (Table 1). Feed and water was provided *ad libitum* and the experiment lasted for 28 days.

Birds in T1 was fed basal diet with Oxytetracycline at 1.2 g/kg feed, T2, T3, T4 and T5 were fed basal diet with Sunflower oil (SFO) at 0.1, 0.2, 0.3 and 0.4 mls/kg feed.

Measurements

Feed intake (g) was estimated from the difference between feed served and left over.

Body weight gain (g/bird) = final weight (g) – initial weight (g)

Average daily gain (ADG/bird) = $\frac{\text{Final body weight} - \text{Initial body weight}}{\text{Total days of the experiment}}$

Average daily feed intake (ADFI/bird) = $\frac{\text{Total feed intake}}{\text{Total days of the experiment}}$

Feed conversion ratio (FCR) = feed intake (g)/weight gain (g)

Mortality was recorded as it occurs.

Oxidative stress indices

On the 28th day of the experiment blood samples were collected via wing web from 6 randomly selected birds per treatment for oxidative stress analysis. Activities of superoxide dismutase (SDA), glutathione peroxidase (GPx), catalase (CAT) and malonyldialdehyde (MLA) were carried out using method outlined by Mahipal *et al.* (2015).

Chemical analysis

Proximate composition of the experimental diet was determined using procedures of AOAC (2000). Quantitative phytochemical determination (saponins, flavonoids, phenolics, alkaloids, steroids and

terpenoids) was carried out according to the method outlined by Harbone (1973); Odebiyi and Sofowora (1978) and Boham and Kocipai (1974).

Statistical analysis

Data collected on performance and nutrient retention were subjected to analysis of variance (ANOVA) using SAS statistical package, SAS (2000). The means were separated using Duncan multiple range test of the same software.

Table 1: Vaccination program for broiler chicks

| Age of birds | Vaccines | Route of administration |
|--------------|-------------------------|-------------------------|
| Day 7 | 1 st Lasota | Oral via drinking water |
| Day 9 | 1 st Gumboro | Oral via drinking water |
| Day 13 | 2 nd Gumboro | Oral via drinking water |
| Day 18 | 2 nd Lasota | Oral via drinking water |

Table 2: Ingredient composition of the experimental diets

| Ingredients | Quantity |
|----------------------------|----------|
| Maize | 55.00 |
| Wheat offal | 2.10 |
| Soya bean meal | 10.00 |
| Groundnut cake | 25.23 |
| Fish meal (72%) | 3.00 |
| Limestone | 1.50 |
| Bone meal | 3.00 |
| Lysine | 0.20 |
| Methionine | 0.25 |
| *Premix | 0.25 |
| Salt | 0.30 |
| Toxin binder | 0.10 |
| Total | 100.0 |
| Determined analysis (% DM) | |
| Crude protein | 23.50 |
| Crude fibre | 3.18 |
| Ether extract | 4.10 |
| Calcium | 1.91 |
| Phosphorus | 0.90 |
| Energy (Kcal/kg) | 2933.7 |

*Premix supplied per kg diet: - vit A, 13,000 I.U; vit E, 5mg; vit D3, 3000I.U, vit K, 3mg; vit B2, 5.5mg; Niacin, 25mg; vit B12, 16mg; choline chloride, 120mg; Mn, 5.2mg; Zn, 25mg; Cu, 2.6g; folic acid, 2mg; Fe, 5g; pantothenic acid, 10mg; biotin, 30.5g; antioxidant, 56mg

Results and discussion

Phytochemicals composition of Sunflower oil (SFO)

Table 3 reveals the phytochemicals composition of sunflower oil (SFO). The sample contains alkaloids (3.86 %), condensed tannins (0.96 %), hydrolysable tannins (9.87%), phenols (7.06 %), terpenoids (5.10 %), steroids (4.47 %), saponins (3.51 %) and flavonoids (10.60%). In order of abundance flavonoids > hydrolyzable tannins > phenols > terpenoids > steroids > alkaloids > saponins

> condensed tannins respectively. The result obtained in this study is in conformity with the values recorded by Chinelo and Ujunwa (2017); Jumuna *et al.* (2014) and Labaran *et al.* (2016). Phytochemicals are natural bioactive compounds that are derived from plants and incorporated into livestock feed to enhance productivity (Gadde *et al.*, 2017; Alagbe *et al.*, 2022). They are also regarded as non-nutritive plant chemicals that have either defensive or degenerative protective properties (Prakash *et al.*, 2012). For instance, flavonoids and phenols have been found to exhibit anti-inflammatory, anti-allergic, antioxidant and antibacterial properties (Oluwafemi *et al.*, 2021; Agubosi *et al.*, 2021). Steroids and saponins are very key fertility agents in animals (Atamgba *et al.*, 2015; Shittu and Alagbe, 2020). Alkaloids have been known to perform antibacterial, antispasmodic and analgesic properties (Ahmad *et al.*, 2013). Tannins have been suggested to be involved with antibacterial and anti-viral activity while tannins and flavonoids are thought to be responsible for antidiarrheal activity (Enzo, 2007; Olafadehan *et al.*, 2021). Terpenoids has high therapeutic value and function as antimicrobial, anticarcinogenic and anti-diuretic (Sittanikove *et al.*, 2001).

Table 3: Phytochemicals composition of Sunflower oil

| Parameters | % Composition |
|----------------------|---------------|
| Alkaloids | 3.86 |
| Condemned tannins | 0.96 |
| Hydrolysable tannins | 9.87 |
| Phenols | 7.06 |
| Terpenoids | 5.10 |
| Steroids | 4.47 |
| Saponins | 3.51 |
| Flavonoids | 10.06 |

Growth performance of broiler chicks fed different dietary inclusion of SFO

Table 4 shows the growth performance of broiler chicks fed different dietary inclusion of SFO. Inclusion levels of sunflower oil in the diets of broiler chicks had significant effect ($P<0.05$) on the average body weight gain (WG), average daily weight gain (ADWG), average daily feed intake (ADFI) and feed conversion ratio (FCR). WG (486.26 – 538.01 g), ADWG (16.51 – 19.21 g), ADFI (39.31 – 40.00 g) and FCR (1.80 – 2.18). Broiler chicks fed SFO at 0.3, 0.4 and 0.5 mL/kg feed had similar body weight gain which is significantly higher than birds fed 0.1 ml of sunflower oil and the control. The higher final body weight observed among birds on 0.2, 0.3 and 0.4 mls of SFO could be attributed to the activities of phytochemicals which have been proven to stimulate functions of the intestinal tract to improve digestive secretions, nutrient absorption and metabolism (Alagbe and Oluwafemi, 2019).

The dietary inclusion of sunflower oil in broiler chicks also exerted a significant ($P<0.05$) difference in the feed intake. It was observed that birds in T1 had the lowest feed intake when compared to the other groups. This is a clear indication that SFO is capable of improving the palatability of feed which further translates to efficient utilization of feed. The result obtained in this study is in agreement with the findings of Amad *et al.* (2011) but contrary to the reports of Halle *et al.* (2004) who recorded a numerical decrease in feed intake of birds fed a blend of thyme, star anise and oregano leaves and its associated essential oil

Birds fed 0.2, 0.3, and 0.4 mls of SFO had an excellent FCR when compared to the other groups ($P<0.05$). This could be attributed to the mechanisms of stimulation of digestive enzyme secretion and the stabilization of the ecosystem of gut microflora, leading to improved feed utilization and less exposure to growth-depressing disorders associated with digestion and metabolism as reported by (Bento *et al.*, 2013; Franz *et al.*, 2010 and Kurekci *et al.*, 2014) which is attributed to essential oil. The

positive effects of essential oils on suppressing the activities of pathogenic microorganisms have been reported in many broiler studies due to the presence of bioactive compounds or phytochemicals (Oluwafemi *et al.*, 2021; Alagbe and Akintayo, 2021). This explains the reason why no mortality was recorded among birds fed with SFO compared to the control.

Table 4: Growth performance of broiler chicks fed different dietary inclusion of SFO

| Parameters | Inclusion levels of sunflower oil (mL) | | | | | SEM | LOS |
|---------------|--|---------------------|----------------------|----------------------|----------------------|-------|-----|
| | 0.0 | 0.1 | 0.2 | 0.3 | 0.4 | | |
| IBW (g) | 39.67 | 39.67 | 39.83 | 39.93 | 39.77 | 0.19 | NS |
| FBW (g) | 525.93 ^c | 501.85 ^c | 553.73 ^{ab} | 555.56 ^{ab} | 577.78 ^a | 16.35 | * |
| WG (g) | 486.26 ^c | 462.19 ^c | 513.87 ^{ab} | 515.62 ^{ab} | 538.01 ^a | 16.30 | * |
| ADWG (g) | 17.37 ^{bc} | 16.51 ^c | 18.35 ^{ab} | 18.42 ^{ab} | 19.21 ^a | 0.58 | * |
| FI (g) | 1100.70 | 1109.37 | 1139.19 | 1123.50 ^b | 1120.04 ^b | 5.39 | * |
| ADFI (g) | 39.31 ^c | 39.62 ^c | 40.69 ^a | 40.13 ^b | 40.00 ^b | 0.19 | * |
| FCR | 2.18 ^a | 2.13 ^a | 2.00 ^b | 1.85 ^c | 1.80 ^c | 0.06 | * |
| Mortality (%) | 1.00 | - | - | - | - | 0.01 | * |

^{a-c}Means in the same row with different superscript differ significantly ($P < 0.05$); IBW: initial body weight; FBW: final body weight; WG: weight gain; ADWG: average daily weight gain; FI: feed intake; ADFI: average daily feed intake; SEM: standard error of means; FCR: feed conversion ratio T1: basal diet + 0 % SFO; T2: basal diet + 0.1 % SFO; T3: basal diet + 0.2 % SFO; T4: basal diet + 0.3 % SFO; T5: basal diet + 0.4 % SFO; LOS: level of significance.

Oxidative status of broiler chicks fed different levels of SFO

Oxidative status of broiler chicks fed different levels of SFO is presented in Table 5. Activities of superoxide dismutase (SDA), glutathione peroxidase (GPx), catalase (CAT) and malonyldialdehyde (MLA) values ranged from 23.00 – 34.12 (U/mg Hb), 18.93 – 29.12 (U/mg Hb), 54.21 – 63.88 (U/mg Hb) and 1.96 – 4.08 (U/mg Hb) respectively. SDA, GPx, CAT and MLA values were significantly ($P < 0.05$) influenced by the treatments. MLA, SDA, CAT and GPx values were highest in T1, intermediate in T2, T3 and lowest in T4 and T5 ($P < 0.05$). The values decreases as the level of SFO inclusion increases across the treatment. This shows that SFO is capable of scavenging free radicals, thus preventing diseases in the body of animals. The presence of bioactive compounds like flavonoids and phenols in SFO could help to further boost the immune system and prevent oxidative damage to biomolecules, superoxide anions and lipid peroxy radicals (Hollman, 2001; Alagbe *et al.*, 2021). The result obtained in this study is in agreement with the findings of Oluwafemi *et al.* (2021).

Table 5: Oxidative status of broiler chicks fed different levels of SFO

| Parameters | T1 | T2 | T3 | T4 | T5 | SEM | LOS |
|---------------|--------------------|--------------------|--------------------|--------------------|--------------------|------|-----|
| MLA (U/mg Hb) | 4.08 ^a | 2.12 ^b | 2.08 ^b | 2.00 ^c | 1.96 ^c | 0.09 | * |
| SDA (U/mg Hb) | 34.12 ^a | 30.80 ^b | 28.79 ^b | 23.08 ^c | 23.00 ^c | 2.04 | * |
| GPx (U/mg Hb) | 29.12 ^a | 25.40 ^b | 23.81 ^b | 20.87 ^c | 18.93 ^c | 1.72 | * |
| CAT (U/mg Hb) | 64.21 ^a | 55.77 ^b | 51.51 ^b | 49.60 ^c | 49.88 ^c | 0.32 | * |

Means in the same row with different superscripts differ significantly ($P < 0.05$); superoxide dismutase (SDA); glutathione peroxidase (GPx); catalase (CAT); malonyldialdehyde (MLA)

Conclusion

Sunflower oil contains several phytochemicals capable of promoting food safety without causing any side effect on continuous use and the dietary inclusion of essential oil at 0.4 ml/kg basal diet in broilers

is capable of maintaining good health, suppressing the activities of pathogenic microorganisms and scavenging free radicals.

References

1. Olafadehan, O.A., Oluwafemi, R.A and Alagbe, J.O (2020). Carcass quality, nutrient retention and caeca microbial population of broiler chicks administered Rolfe (*Daniellia oliveri*) leaf extract as an antibiotic alternative. *Journal of Drug Discovery*. 14(33):146-154.
2. A.O.A.C.(2000). Association of Official Analytical Chemists. Official Methods of Analysis 19th Edition Washington, D.C Pages 69-77.
3. Oluwafemi, R.A., Agubosi, O.C.P and Alagbe, J.O. (2021). Proximate, minerals, vitamins and amino acid composition of *Prosopis africana* (African mesquite) seed oil. *Asian Journal of Advances in Research* 11(1): 21-27.
4. Oluwafemi, R.A., Uankhoba, I.P and Alagbe, J.O. (2021). Effects of turmeric oil as a dietary supplements on the growth performance and carcass characteristics of broiler chicken. *International Journal of Orange Technologies*, 3(4): 1-9.
5. Olafadehan, O.A., Oluwafemi, R.A and Alagbe, J.O. (2020). Performance, haemato-biochemical parameters of broiler chicks administered Rolfe (*Daniellia oliveri*) leaf extract as an antibiotic alternative. *Advances in Research and Reviews*, 2020, 1:4.
6. Oluwafemi, R.A., Isiaka Olawale and Alagbe, J.O. (2020). Recent trends in the utilization of medicinal plants as growth promoters in poultry nutrition- A review. *Research in: Agricultural and Veterinary Sciences*. 4(1): 5-11.
7. Mahipal, C., Ashak, K.P., Shalini, B., Narayan, D., Sunil, E.J and Kusumakar, S. (2015). Dietary supplementation of a novel phyto-genic feed additive: effects on nutrient metabolism, antioxidant status and immune response of goats. *Animal Production Science*, 2:21-28. <http://dx.doi.org/10.1071/AN14770>.
8. Alagbe, J.O. (2019). Haematology, serum biochemistry, relative organ weight and bacteria count of broiler chicken given different levels of *Luffa aegyptiaca* leaf extracts. *International Journal of Advanced Biological and Biomedical Research*. 7(4):382-392.
9. Omokore, E.O and Alagbe, J.O. (2019). Efficacy of dried *Phyllanthus amarus* leaf meal as an herbal feed additive on the growth performance, haematology and serum biochemistry of growing rabbits. *International Journal of Academic Research and Development*. 4(3): 97-104.
10. Alagbe, J.O., Shittu, M.D and Eunice Abidemi Ojo (2020). Prospect of leaf extracts on the performance and blood profile of monogastric – A review. *International Journal of Integrated Education*. 3(7): 122-127.
11. Alagbe, J.O and Oluwafemi, R.A. (2019). Growth performance of weaner rabbits fed Noni (*Morinda citrifolia*) and *Moringa olifera* leaf mixture as partial replacement of soya bean meal. *International Journal of Advanced Biological and Biomedical Research*. 7(2): 185-195
12. Aduku, A. O. (1994). Nutrient requirement of chicken. Feed ingredient analysis table and their uses. In: *Revamping poultry production, feeding and management training manual*. Edited by S. O. Ogundipe, O. O. Oni and A. A. Sekoni, NSPRI/ ABU Shika- Zaria.
13. Adewale, A.O., Alagbe, J.O., Adeoye, Adekemi. O. (2021). Dietary Supplementation of *Rauvolfia Vomitoria* Root Extract as A Phyto-genic Feed Additive in Growing Rabbit Diets: Haematology and serum biochemical indices. *International Journal of Orange Technologies*, 3(3): 1-12.

14. Agubosi, O.C.P., Oluwafemi, R.A., and Alagbe, J.O. (2021). Preliminary study on GC-MS analysis of *Prosopis africana* seed (African mesquite) oil. *Journal of Ethics and Diversity in International Communication* 1(4): 18-20.
15. Agubosi, O.C.P., Oluwafemi, R.A and Alagbe, J.O. (2021). The effect of processing on the proximate, mineral and vitamin composition of Neem leaves (*Azadirachta indica*) grown in Gwagwalada, FCT, Abuja. *Abuja Journal of Agriculture and Environment*, 1(1): 293-299.
16. Shittu, M.D and Alagbe, J.O. (2020). Phyto-nutritional profiles of broom weed (*Sida acuta*) leaf extract. *International Journal of Integrated Education*. 3(11): 119-124.
17. Alagbe, J.O., Shittu, M.D and Tanimomo, Babatunde K. (2022). Influence of *Anogeissus leio carpus* stem bark on the fatty acid composition in meat of broiler chickens. *European Journal of Life Safety and Stability* 14(22): 13-22.
18. Agubosi, O.C.P., Imudia, Favour Dumkenechukwu and Alagbe, J.O. (2022). Evaluation of the nutritional value of air dried and sun-dried sweet potato (*Ipomoea batatas*) peels. *European Journal of Life Safety and Stability* 14(22): 43-51.
19. Chinelo, A.E and Ujunwa, C.N. (2017). Investigation of phytochemical and proximate components in different parts of *Boerhavia diffusa* and *B. erecta* L. *Advances in Sciences*, 2(5):60-63
20. Chiasson H., Belanger A, Bostanian N. 2001. Acaridial properties of *Asteris absinthium* and *Tanacetum vulgare* (Asteraceae) essential oils obtained by three methods of extraction. *Applied Environment Microbial*. 47:229-233
21. WHO (World Health Organization) (2014). Antimicrobial Resistance: Global Report on Surveillance 2014. Accessed June 2019.
22. Windisch W, Schedle K, Plitzner C, Kroismayr A. (2008). Use of phytogenic products as feed additives for swine and poultry. *Journal of Animal Science* 86:140-8.
23. Franz C, Baser K, Windisch W. (2010). Essential oils and aromatic plants in animal feeding a European perspective. A review. *Flavour Fragr Journal* 25:327-40.
24. Bento M, Ouwehand A, Tiihonen K, Lahtinen S, Nurminen P, Saarinen M. (2013). Essential oils and their use in animal feeds for monogastric animals-Effects on feed quality, gut microbiota, growth performance and food safety: a review. *Journal of Veterinary Medicine* 58:449-58.
25. Amad A, Maanner K, Wendler K, Neumann K, Zentek J.(2011). Effects of a phytogenic feed additive on growth performance and ileal nutrient digestibility in broiler chickens. *Poultry Science* 90:2811-6.
26. Jamuna S, Paulsamy S, Karthika K. (2014). *In vitro* antifungal activity of leaf and root extracts of the medicinal plant, *Hypochaeris radicata* L. *International Journal Pharmaceutical Science* 5(3): 758-761.
27. Labaran, I., Lukman, O.A., Adam, J.D and Usman, M. (2016). Analysis of some phytochemicals and minerals found in aqueous stem bark of *Albizia lebbek*. *Dutse Journal of Pure and Applied Science*, 2(1): 231-237.
28. Sittanikove, Y.X, Yeo, C.R, Chung HL, Yuk, H.G. (2001). Plant essential oils as active antimicrobial agents. *Critical Review Food Science and Nutrition* 54:625-44.
29. Atamgba, A. A., Margret, A. A., Kayode, D. and Amonor, J. W. (2015). The biomedical significance of the phytochemical, proximate and mineral composition of the leaf, stem bark and roots of *Jatropha curcas*. *Asian Pacific Journal of Tropical Biomedicine*, 5(8):650-657

30. Enzo, A. P. (2007). Traditional plants and herbal remedies used in the treatment of diarrheal disease: Mode of action, quality, efficacy and safety considerations. In: Ahmad, I., Aqil, F. and Owais, M. editors *Modern Phytomedicine Turning Medicinal Plants in to Drugs*, 248-260.
31. Hollman, P.C. (2001). Evidence for health benefits of plant phenols: Local or systemic effects. *Journal of Food science and Agriculture*, 81: 842-852.
32. Halle, I., Thomann, R., Bauermann, U., Henning, M. and Kohler, P. (2004). Effects of a graded supplementation of herbs and essential oils in broiler feed on growth and carcass traits. *Landbauforschung Volkenrod*, 54:219-229.
33. Gadde, F., Madrid, J., Garcia, V., Orengo, J. and Megias, M. (2017). Influence of two plants extracts on broilers performance, digestibility, and digestive organ size. *Poultry science*, 83:169-174.
Boham, B. A. and Kocipai, A. C. (1974). Flavonoids and condensed tannins from leaves of Hawaiian vaccinium vaticulatum and V. calycinium. *Pacific Sci.* 48: 458-463.
34. Harborne, J. D. (1973). *Phytochemical methods: A guide to modern techniques of plant analysis*. Chapman and Hall, London. 279.
35. Odebiyi, A. and Sofowora, A. E. (1978). *Phytochemical Screening of Nigerian Medicinal Plant*. Part III, *Lloydia*, 41, 234- 246.

